

ENGELHARDT ET AL., U.S. PAT. APPL. SER. NO. 08/486,069
Consolidated Claims [As Of November 20, 2001]
Exhibit 1 To Applicants' November 20, 2001 Consolidation Amendment

569. (Twice Amended) A process for determining the sequence of a nucleic acid of interest, comprising the steps of:

providing or generating detectable non-radioactively labeled nucleic acid fragments, each fragment comprising a sequence complementary to said nucleic acid of interest or to a portion thereof, wherein each of said fragments comprises one or more detectable non-radioactively modified or labeled nucleotides or nucleotide analogs, which nucleotide analogs can be attached to or coupled to or incorporated into DNA or RNA, and wherein said one or more detectable non-radioactively modified or labeled nucleotides or nucleotide analogs have been modified or labeled on at least one of the sugar moiety, the sugar analog, the phosphate moiety, the phosphate analog, the base moiety, or the base analog thereof;

subjecting said detectable non-radioactively labeled fragments to a sequencing gel to separate or resolve said fragments; and

detecting non-radioactively the presence of each of said separated or resolved fragments by means of said detectable non-radioactively modified or labeled nucleotides or nucleotide analogs, and determining the sequence of said nucleic acid of interest.

570. (NEW) The process according to claim 569, wherein the nucleic acid sequence of interest is derived from an organism.

571. (NEW) The process according to claim 570, wherein said organism is selected from the group consisting of bacteria, fungi, viruses, yeast, mammals, and a combination of any of the foregoing.

721. (Twice Amended) A process for determining the sequence of a nucleic acid of interest, comprising the steps of:

providing or generating detectable non-radioactively labeled nucleic acid fragments, each fragment comprising a sequence complementary to said nucleic acid of interest or to a portion thereof, wherein each of said fragments comprises one or more detectable non-radioactively modified or labeled nucleotides or nucleotide analogs, which nucleotide analogs can be attached to or coupled to or incorporated into DNA or RNA, and wherein said one or more detectable non-radioactively modified or labeled nucleotides or nucleotide analogs have been modified or labeled on at least one of the sugar moiety, the sugar analog, the phosphate moiety, the phosphate analog, the base moiety, or the base analog thereof;

introducing or subjecting said detectable non-radioactively labeled fragments to a sequencing gel;

separating or resolving said fragments in said sequencing gel; and detecting non-radioactively each of the separated or resolved fragments; and determining the sequence of said nucleic acid of interest.

722. (NEW) The process according to claim 721, wherein the nucleic acid sequence of interest is derived from an organism.

723. (Amended) The process according to claims 722 or 726, wherein said organism is selected from the group consisting of bacteria, fungi, viruses, yeast, mammals, and a combination of any of the foregoing.

724. (NEW) The process according to claim 723, wherein said organism comprises a mammal.

873. (Twice Amended) A process for determining the sequence of a nucleic acid of interest, comprising the steps of:

providing or generating detectable non-radioactive labeled nucleic acid fragments, each fragment comprising a sequence complementary to said nucleic acid of interest or to a portion thereof, wherein each of said fragments comprises one or more detectable non-radioactive modified or labeled nucleotides or nucleotide analogs, which nucleotide analogs can be attached to or coupled to or incorporated into DNA or RNA, and wherein said one or more detectable non-radioactive modified or labeled nucleotides or nucleotide analogs have been modified or labeled on at least one of the sugar moiety, the sugar analog, the phosphate moiety, the phosphate analog, the base moiety or the base analog thereof;

detecting non-radioactively the detectable non-radioactive labeled nucleic acid fragments with a sequencing gel; and

determining the sequence of said nucleic acid of interest.

874. (NEW) The process according to claim 873, wherein the nucleic acid sequence of interest is derived from an organism.

875. (NEW) The process according to claim 874, wherein said organism is selected from the group consisting of bacteria, fungi, viruses, yeast, mammals, and a combination of any of the foregoing.

876. (Amended) The process according to claims 875 or 878, wherein said organism comprises a mammal.

877. (NEW) The process according to claim 876, wherein said mammal comprises a human being.

1025. (Twice Amended) A process for determining the sequence of a nucleic acid of interest, comprising the step of detecting non-radioactively with a sequencing gel one or more detectable non-radioactive labeled nucleic acid fragments comprising a sequence complementary to said nucleic acid of interest or to a portion thereof, wherein each of said fragments comprises one or more detectable non-radioactive modified or labeled nucleotides or nucleotide analogs, which nucleotide analogs can be attached to or coupled to or incorporated into DNA or RNA, and wherein said one or more detectable non-radioactive modified or labeled nucleotides or nucleotide analogs have been modified on at least one of the sugar moiety, the sugar analog, the phosphate moiety, the base moiety or the base analog thereof.

1026. (NEW) The process according to claim 1025, wherein the nucleic acid sequence of interest is derived from an organism.

1027. (Amended) The process according to claims 1026 or 1030, wherein said organism is selected from the group consisting of bacteria, fungi, viruses, yeast, mammals, and a combination of any of the foregoing.

1028. (NEW) The process according to claim 1027, wherein said organism comprises a mammal.

1029. (NEW) The process according to claim 1028, wherein said mammal comprises a human being.

1030. (NEW) The process according to claim 1026, wherein said organism is living.

1573. (Amended) The process according to claim 1475, wherein each of said set of clones or DNA fragments or oligo- or polynucleotides is labeled with the same indicator molecule.

1574. (NEW) The process according to any of claims. 1473, 1474 or 1475, wherein said detecting step is carried out by a means selected from the group consisting of manual means and automatic means.

1575. (NEW) The process according to claim 1574, wherein said manual means comprises visualization.

1576. (NEW) The process according to claim 1574, wherein said automatic means comprises computerized automatic karyotyping.

1577. (NEW) The process according to claim 1476, wherein each of said sets of clones or DNA fragments or oligo- or polynucleotides is labeled with the same indicator molecule.

1578. (NEW) The process according to claim 1476, wherein each of said sets of clones or DNA fragments or oligo- or polynucleotides is labeled with a different indicator molecule.

1579. (NEW) The process according to claim 1476, wherein said detecting and determining step is carried out by a means selected from the group consisting of manual means and automatic means.

1580. (NEW) The process according to claim 1579, wherein said manual means comprises visualization.

1476. (Amended) A process for determining the number of chromosomes in an interphase cell of interest, the process comprising the steps of:

providing sets of clones or DNA fragments or oligo- or polynucleotides derived from said clones, wherein said set of clones or DNA fragments or oligo- or polynucleotides are specifically complementary to or specifically hybridizable with at least one locus or loci in a chromosome of said interphase cell of interest and each of said clones or DNA fragments or oligo- or polynucleotides in said sets comprises one or more detectable non-radioactive modified or labeled nucleotides or nucleotide analogs, which nucleotide analogs can be attached to or coupled to or incorporated into DNA or RNA, and wherein said detectable non-radioactive modified or labeled nucleotides or nucleotide analogs are selected from the group consisting of:

(i) a nucleotide or nucleotide analog having the formula

PM—SM—BASE—Sig

wherein

PM is a phosphate moiety or phosphate analog,

SM is a sugar moiety or sugar analog,

BASE is a pyrimidine, a purine, or a 7-deazapurine base moiety, or a base analog of any of the foregoing, and

Sig is a detectable non-radioactive moiety,

wherein PM is covalently attached to SM, BASE is covalently attached to SM, and Sig is covalently attached to BASE at a position other than the C5 position when BASE is a pyrimidine moiety or a pyrimidine analog, at a position other than the C8

position when BASE is a purine or a purine analog, and at a position other than the C7 position when BASE is a 7-deazapurine or a 7-deazapurine analog;

(ii) a nucleotide or nucleotide analog having the formula

Sig

PM—SM—BASE

wherein

PM is a phosphate moiety or phosphate analog,

SM is a sugar moiety or sugar analog,

BASE is a pyrimidine, a purine or a 7-deazapurine base moiety, or a base analog of any of the foregoing, and

Sig is a detectable non-radioactive moiety,

wherein PM is covalently attached to SM, BASE is covalently attached to SM, and

Sig is covalently attached SM directly or through a linkage group; and

(iii) a nucleotide or nucleotide analog, said nucleotide having the formula

Sig—PM—SM—BASE

wherein

PM is a phosphate moiety or phosphate analog,

SM is a sugar moiety or sugar analog,

BASE is a pyrimidine, a purine or a 7-deazapurine base moiety, or a base analog of any of the foregoing, and

Sig is detectable non-radioactive moiety,

wherein PM is covalently attached to the SM, BASE is covalently attached to SM, and Sig is covalently attached to PM directly or through a linkage group;

contacting said interphase cell under hybridizing conditions with said sets of clones or DNA fragments or oligo- or polynucleotides, and permitting specific hybridization of said sets of clones or DNA fragments or oligo- or polynucleotides to any of the locus or loci in said chromosomes;

detecting non-radioactively any of said sets of clones or DNA fragments or oligo- or polynucleotides specifically hybridized to the locus or loci in said chromosomes, to obtain a pattern of generated signals; and comparing each generated signal with other generated signals in said pattern, and determining the number of chromosomes in said interphase cell of interest.

1477. (NEW) The process according to any of claims 1473, 1474, 1475 or 1476, wherein said nucleotide analog can be attached terminally to DNA or RNA by means of an enzyme.

1478. (NEW) The process according to claim 1477, wherein said enzyme comprises terminal transferase.

1479. (NEW) The process according to any of claims 1473, 1474, 1475 or 1476, wherein said nucleotide analog can be coupled to DNA or RNA by a coupling means selected from the group consisting of chemical coupling and enzymatic coupling.

1480. (Wholly Rewritten) The process according to claim 1479, wherein said chemical coupling can be carried out by a chemical coupling means selected from the group consisting of carbodiimide and formaldehyde.